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Occurrence of *Haemoproteus nettionis* in Wood Ducks (*Aix sponsa* L.)[1]

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Abstract. The protozoan *Haemoproteus nettionis* was found in 77 of 168 wood ducks (*Aix sponsa* L.) live-trapped on the Upper Mississippi River Wildlife and Fish Refuge in September, 1963. No parasitemia was found in 68 wood ducks live-trapped in July and August, 1959 at the Union Slough National Wildlife Refuge in north-central Iowa.

A description of the parasite from wood ducks on the Upper Mississippi Refuge is presented.

Haemoproteus was first reported from Anatidae by Johnston and Cleland, (1909) in the Australian teal (*Anas (nettion) castanea*). They named the parasite *Halteridium nettionis*, which was later recognized as *Haemoproteus nettionis*. Since 1909 *Haemoproteus* has been reported from 25 species of waterfowl. Coatney (1936), Levine and Hanson (1953), and Herman (1954) reviewed most of the records of *Haemoproteus* from waterfowl.

Nelson and Gashwiler (1941) first reported *Haemoproteus* from wood ducks (*Aix sponsa* L.) in Maine. Herman (1934) reported a similar parasitemia in wood ducks from Maryland.

Herman (1954) suggested *Haemoproteus nettionis* (Johnston and Cleland, 1909) Coatney, 1936 be accepted as the correct name for *Haemoproteus* in North American Anatidae. This classification was followed in this paper. Herman (1951) listed synonyms for *Haemoproteus* in waterfowl and an amended description of the parasite. Levine and Hanson (1953) discussed the morphological characteristics of *Haemoproteus* in several species of waterfowl.

Comparisons of *Haemoproteus* from various waterfowl species have been based on morphological information, and few data are available on cross-transmission experiments. Additional studies are needed to determine if morphological differences found by several authors are valid criteria for separating *Haemoproteus* in Anatidae into more than one species. Halba (1946) named a parasite from the black duck (*Anas rubripes*)

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Haemoproteus anatis. Levine and Hanson (1953) choose the same name for a parasite of the Canada goose (*Branta canadensis interior*). They based their identification on morphological differences with described types, and cross-transmission experiments were not conducted. Levine and Hanson agreed that *Haemoproteus nettionis* is the proper name for *Haemoproteus* in the Anatidae if *Haemoproteus anatis* is cross transmissible (Herman, 1951). The parasite found in this study was not morphologically similar to all the various descriptions of waterfowl *Haemoproteus* in the literature; however, cross-transmission experiments were not performed. Thus, it was logical to identify the parasite as *Haemoproteus nettionis* until new evidence indicates it might be another species.

The occurrence of *Haemoproteus* in wood ducks in Iowa was unknown prior to this study. Farmer (1960) and Roslien (1963) found *Haemoproteus* in non-game species in Iowa, and it was assumed conditions were suitable for the parasite. This study was undertaken to determine the prevalence of *Haemoproteus* and other blood parasites in selected wood duck populations.

Study Areas

Blood smears were prepared from wood ducks at three locations. Smears from 168 ducks on pools 9 and 10 of the Upper Mississippi River Wildlife and Fish Refuge in northeast Iowa were prepared in September, 1963. Blood smears from 68 ducks on the Union Slough National Wildlife Refuge in northcentral Iowa (Kossuth County) were prepared in July and August, 1959. The Upper Mississippi River Refuge supported large numbers of wood ducks during summer and autumn while the Union Slough Refuge supported fewer wood ducks during these breeding and migration periods.

Materials And Methods

Peripheral-blood smears were prepared from wood ducks live-trapped by refuge personnel operating seasonal banding traps. Blood drawn from the brachial vein was used to make the smears as recommended by McClure and Cedeno (1955). The smears were air-dried and fixed in methyl alcohol for 1 minute before being stained in unbuffered Giemsas' stain (1:40 for 30 minutes).

An A. O. Spencer binocular microscope with 10X oculars and a 10X objective was used for preliminary examination of blood smears. Erythrocytes which appeared parasitized under low power (100X) were inspected under oil immerson (970X). If a smear appeared negative under low power, it was examined under oil immersion for 10 minutes. Smears containing parasitized cells were usually located after 1 or 2 minutes of seaching at

low power. All smears were examined for at least 10 minutes with observations of from 250-350 different fields. *Haemoproteus nettionis* was characterized under oil immersion and specific identification accomplished by making comparisons with published descriptions.

Results And Discussion

Haemoproteus Prevalence. Blood smears prepared from 168 wood ducks captured on the Upper Mississippi River Refuge revealed 77 positive for *Haemoproteus* (Table 1). Parasitemias occurred in 43% of 21 adults and in 48% of 147 immatures. Blood smears prepared from 68 wood ducks at Union Slough Refuge were negative for protozoan blood parasites (Table 2).

Table 1. Prevalence of *Haemoproteus nettionis* in wood ducks trapped on the Upper Mississippi River Refuge, September, 1960.

		Sample		Incidence	
Age	Sex	Number examined	Per cent of sample	Number positive	Per cent positive
Adult	Male	13	8	7	54
Adult	Female	8	5	2	25
Immature	Male	84	50	38	45
Immature	Female	63	37	30	48
		168	100	77	Avg. 46

Table 2. Prevalence of *Haemoproteus nettionis* in wood ducks trapped on the Union Slough Refuge, July and August 1959.

		Sample		Incidence	
Age	Sex	Number examined	Per cent of sample	Number positive	Per cent positive
Adult	Male	14	21	0	0
Adult	Female	3	4	0	0
Immature	Male	39	57	0	0
Immature	Female	12	18	0	0
		68	100	0	0

Haemoproteus Description. Smears prepared from peripheral blood revealed sexual stages of *Haemoproteus* in the erythrocytes of the wood duck. Mature gametocytes were observed often and young stages were rare. The absence of young stages was anticipated since the smears were prepared in September and peak transmission probably occurred in July and August.

Gametocytes usually occupied cytoplasm of the host cell except for occasional free forms which were rounded. Microgametocytes appeared bluish-pink when stained with Giemsas stain (1:40 for 30 Minutes). Nuclear material stained pink and was diffused throughout the parasite. Macrogametocytes stained light blue and showed alveolar cytoplasm. Nuclear material in the female stained red and was located in a compact oblong mass in the center of the parasite.

The parasite usually did not enlarge the host erythrocyte or displace the host cell nucleous (Figure 1). Occasionally the host cell appeared elongate, or its nucleous was displaced by the gametocyte (Figure 1). Many forms of waterfowl *Haemoproteus* have been reported to laterally displace the nucleous of the host cell. This was observed by Levine and Hanson (1953) in the Canada goose, by Johnston and Cleland (1909) in Australian teal, and by Herman (1938, 1951) in the black duck (*Anas rubripes*) and wood duck respectively.

FIGURE 1. Red blood cells of a wood duck showing two cells parasitized with *Haemoproteus nettionis*; "a" is an infected cell as it usually appears, and "b" shows a rare form with the nucleus to one side.

Wetmore (1911) and Haiba (1948, 1948) reported similar displacement of the host nucleous. The absence of nuclear displacement observed in this study was also noted by Leger (1918) working with the Muscovy duck (*Cairina* (*Anas*) *moschata*), which is closely related to the wood duck. Nelson and Gashwiler (1941) observed *Haemoproteus* in the wood duck, but failed to mention parasite morphology.

Some parasitized cells displayed a narrow irregular band of cytoplasm between the parasite cell membrane and the nucleus of the host cell. Herman (1951) and Levine and Hanson (1953) observed a similar characteristic. The cell membrane of the parasite was in close contact with the erythrocyte cell membrane over at least one-half of its periphery. Wetmore (1911) reported that *Haemoproteus* gametocytes in the common mallard (*Anas p. platyrhynchos*) were in contact with the host cell periphery. Mature gametocytes usually filled the entire host cell cytoplasm. An occasional gametocyte did not fill a small area of cytoplasm on one side of the host cell nucleous.

Pigment granules were numerous in most gametocytes with a mean number of 37 (27-51) per parasite. The granules were usually round and varied from small to moderate in size. Large granules were not observed. The pigment granules appeared rod-shaped in a few parasites. Granules were usually numerous near the poles of the parasite. Gametocytes with uniform distribution of granules in the cytoplasm were seldom observed.

Johnston and Clelands' (1909) original description of *Haemoproteus* in Australian teal indicated 13-30 pigment granules distributed more or less uniformly in the cytoplasm with a tendency to be more numerous at the poles. Levine and Hanson (1953) reported *Haemoproteus* in the Canada goose contained 16 to more than 50 medium-sized round pigment granules distributed throughout the cytoplasm with a tendency to be more numerous at the ends. Leger (1918) reported numerous pigment granules in *Haemoproteus* from the Muscovy duck. *Haemoproteus* from the white-winged duck (*Asarcornis scutulata*) contained 50-60 pigment granules per gametocyte (Kowarski *et al.*, 1937). The number of pigment granules appears to vary in different hosts, although other morphological features are similar.

Gametocytes averaged 14.0 microns, and mature gametocytes ranged from 12.4 to 16.1 microns in length. Parasitemias ranged from one infected erythrocyte per 100 fields (970X) to two infected erythrocytes per single oil immerson field.

Haemoproteus Transmission. Herman (1951) succeeded in transmitting *Haemoproteus nettionis* from wood ducks to Indian runner ducks (*Anas platyrhynchos*). He concluded the vector was probably a free-flying insect since transmission occurred away from water as well as near water. Fallis and Wood (1957) suggest *Haemoproteus* in waterfowl is transmitted by biting midges (*Ceratopogonidae*).

Herman (1951) reported the duration of parasitemia as 7 to 12 days. Such a duration indicated that transmission on the Upper Mississippi River Refuge probably occurred during late July or August. Studies of transmission of *Haemoproteus* in northeast Iowa must be completed before it can be determined if the vector(s) are *Ceratopogonidae* or the traditional *Haemoproteus* vector *Hippoboscidae*. No authentic reports of hippoboscid flies on Anatidae occurred in the literature (Bequaert, 1953).

Haemoproteus Absence. Lack of parasitemia in wood ducks captured at Union Slough Refuge in July and August suggested the absence of some element in the life cycle of *Haemoproteus* on the area during the study period. The vector potential at Union Slough is far less than at Pools 9 and 10 of the Upper Mississippi River, and this might explain the lack of parasitized ducks. Fewer wood ducks at Union Slough may have provided an insufficient reservoir potential for transmission to occur. Further studies on the two refuges probably would determine the reasons for variations in parasitemia.

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